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Draft Genome Sequences of Various Bacterial Phyla Isolated from the International Space Station

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ABSTRACT

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ANNOUNCEMENT

The International Space Station (ISS) is currently the only long-term human habitat in space. Microgravity disrupts human immune function (1), and close monitoring of the ISS microbiome for increased pathogenicity is thus an ongoing critical task. Here, we report the draft genomes of 96 bacterial strains that were isolated from the ISS (Table 1). Eleven of the 14 species found are common members of the human microbiome, and most can act as opportunistic human pathogens.

TABLE 1.

Accession numbers, sampling locations, and assembly details for bacterial strains isolated from the ISS

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F8_7S_12B	*Acinetobacter pittii*	JAFDRK000000000	SRR13530731	F8	Lab 3 overhead	R2A, 25	66
F8_7S_13B	*Acinetobacter pittii*	JAFDRL000000000	SRR13530730	F8	Lab 3 overhead	R2A, 25	71
F8_7S_14B	*Acinetobacter pittii*	JAFDRM000000000	SRR13530729	F8	Lab 3 overhead	R2A, 25	68
F8_7S_15B	*Acinetobacter pittii*	JAFDRN000000000	SRR13530728	F8	Lab 3 overhead	R2A, 25	70
F8_7S_16B	*Acinetobacter pittii*	JAFDRO000000000	SRR13530727	F8	Lab 3 overhead	R2A, 25	67
F8_7S_17B	*Acinetobacter pittii*	JAFDRP000000000	SRR13530725	F8	Lab 3 overhead	R2A, 25	69
F8_7S_18B	*Acinetobacter pittii*	JAFDRQ000000000	SRR13530724	F8	Lab 3 overhead	R2A, 25	69
F8_7S_4B	*Acinetobacter pittii*	JAFDRX000000000	SRR13530717	F8	Lab 3 overhead	R2A, 25	69
F8_7S_5B	*Acinetobacter pittii*	JAFDRY000000000	SRR13530716	F8	Lab 3 overhead	R2A, 25	66
F8_7S_6P	*Acinetobacter pittii*	JAFDSA000000000	SRR13530713	F8	Lab 3 overhead	BA, 37	67
F8_7S_7B	*Acinetobacter pittii*	JAFDSB000000000	SRR13530712	F8	Lab 3 overhead	R2A, 25	70
F8_8S_11B	*Acinetobacter pittii*	JAFDSE000000000	SRR13530709	F8	Crew quarters	R2A, 25	70

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F8_8S_12B	*Acinetobacter pittii*	JAFDSF000000000	SRR13530708	F8	Crew quarters	R2A, 25	71
F8_8S_2P	*Acinetobacter pittii*	JAFDSJ000000000	SRR13530703	F8	Crew quarters	BA, 37	70
F8_8S_6P	*Acinetobacter pittii*	JAFDSL000000000	SRR13530701	F8	Crew quarters	BA, 37	71
F8_2S_1P	*Cytobacillus horneckiae*	JAFDQP000000000	SRR13530754	F8	WHC	BA, 37	44
F5_7S_P6	*Kocuria indica*	JAFDPQ000000000	SRR13530782	F5	Lab 3 overhead	BA, 37	28
F6_3S_P_1B	*Kocuria indica*	JAFDPV000000000	SRR13530776	F6	ARED	BA, 37	27
F5_7S_P11C	*Kocuria palustris*	JAFDPC000000000	SRR13530748	F5	Lab 3 overhead	BA, 37	36
F5_7S_P2A	*Kocuria palustris*	JAFDPI000000000	SRR13530790	F5	Lab 3 overhead	BA, 37	33
F5_7S_P2B	*Kocuria palustris*	JAFDPJ000000000	SRR13530789	F5	Lab 3 overhead	BA, 37	32
F5_7S_P7	*Kocuria palustris*	JAFDPR000000000	SRR13530780	F5	Lab 3 overhead	BA, 37	32
F5_7S_P8	*Kocuria palustris*	JAFDPS000000000	SRR13530779	F5	Lab 3 overhead	BA, 37	49
F6_1S_P_2	*Kocuria palustris*	JAFDPT000000000	SRR13530778	F6	Cupola	BA, 37	31
F6_7S_B_1	*Kocuria palustris*	JAFDQD000000000	SRR13530767	F6	Lab 3 overhead	R2A, 25	34
F4_5S_F1_F	*Methylobacterium organophilum*	JAFDOX000000000	SRR13530793	F4	Overhead 4	PDA, 25	219
F5_7S_P10B	*Micrococcus luteus*	JAFDOZ000000000	SRR13530781	F5	Lab 3 overhead	BA, 37	309
F5_7S_P11A	*Micrococcus luteus*	JAFDPA000000000	SRR13530770	F5	Lab 3 overhead	BA, 37	249

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F5_7S_P1A	*Micrococcus luteus*	JAFDPG000000000	SRR13530704	F5	Lab 3 overhead	BA, 37	289
F5_7S_P1B	*Micrococcus luteus*	JAFDPH000000000	SRR13530791	F5	Lab 3 overhead	BA, 37	163
F5_7S_P2C	*Micrococcus luteus*	JAFDPK000000000	SRR13530788	F5	Lab 3 overhead	BA, 37	274
F5_7S_P3	*Micrococcus luteus*	JAFDPL000000000	SRR13530787	F5	Lab 3 overhead	BA, 37	534
F6_3S_P_1A	*Micrococcus luteus*	JAFDPU000000000	SRR13530777	F6	ARED	BA, 37	68
F6_7S_P_2	*Micrococcus luteus*	JAFDQF000000000	SRR13530765	F6	Lab 3 overhead	BA, 37	103
F6_3S_P_6	*Pseudoclavibacter alba*	JAFDPW000000000	SRR13530775	F6	ARED	BA, 37	8
F8_1S_1P	*Pseudomonas fulva*	JAFDQI000000000	SRR13530762	F8	Cupola	BA, 37	52
F8_1S_2P	*Pseudomonas fulva*	JAFDQJ000000000	SRR13530761	F8	Cupola	BA, 37	49
F8_1S_3P	*Pseudomonas fulva*	JAFDQK000000000	SRR13530760	F8	Cupola	BA, 37	45
F8_1S_4B	*Pseudomonas fulva*	JAFDQL000000000	SRR13530758	F8	Cupola	R2A, 25	50
F8_1S_5B	*Pseudomonas fulva*	JAFDQM000000000	SRR13530757	F8	Cupola	R2A, 25	50
F8_1S_6B	*Pseudomonas fulva*	JAFDQN000000000	SRR13530756	F8	Cupola	R2A, 25	51
F8_2S_1B	*Pseudomonas fulva*	JAFDQO000000000	SRR13530755	F8	WHC	R2A, 25	51
F8_2S_2P	*Pseudomonas fulva*	JAFDQQ000000000	SRR13530753	F8	WHC	BA, 37	51

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F8_2S_3P	*Pseudomonas fulva*	JAFDQR000000000	SRR13530752	F8	WHC	BA, 37	51
F8_4S_1B	*Pseudomonas fulva*	JAFDQS000000000	SRR13530751	F8	Dining table	R2A, 25	47
F8_5S_16B	*Pseudomonas fulva*	JAFDQT000000000	SRR13530750	F8	Overhead 4	R2A, 25	46
F8_6S_10B	*Pseudomonas fulva*	JAFDQU000000000	SRR13530749	F8	PMM port 1	R2A, 25	43
F8_6S_11B	*Pseudomonas fulva*	JAFDQV000000000	SRR13530747	F8	PMM port 1	R2A, 25	45
F8_6S_12B	*Pseudomonas fulva*	JAFDQW000000000	SRR13530746	F8	PMM port 1	R2A, 25	47
F8_6S_13B	*Pseudomonas fulva*	JAFDQX000000000	SRR13530745	F8	PMM port 1	R2A, 25	46
F8_6S_14B	*Pseudomonas fulva*	JAFDQY000000000	SRR13530744	F8	PMM port 1	R2A, 25	165
F8_6S_15B	*Pseudomonas fulva*	JAFDQZ000000000	SRR13530743	F8	PMM port 1	R2A, 25	45
F8_6S_1P	*Pseudomonas fulva*	JAFDRA000000000	SRR13530742	F8	PMM port 1	BA, 37	47
F8_6S_3B	*Pseudomonas fulva*	JAFDRB000000000	SRR13530741	F8	PMM port 1	R2A, 25	45
F8_6S_3P	*Pseudomonas fulva*	JAFDRC000000000	SRR13530740	F8	PMM port 1	BA, 37	45
F8_6S_4B	*Pseudomonas fulva*	JAFDRD000000000	SRR13530739	F8	PMM port 1	R2A, 25	47
F8_6S_5B	*Pseudomonas fulva*	JAFDRE000000000	SRR13530738	F8	PMM port 1	R2A, 25	44
F8_6S_7B	*Pseudomonas fulva*	JAFDRF000000000	SRR13530736	F8	PMM port 1	R2A, 25	48

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F8_6S_8B	*Pseudomonas fulva*	JAFDRG000000000	SRR13530735	F8	PMM port 1	R2A, 25	48
F8_6S_9B	*Pseudomonas fulva*	JAFDRH000000000	SRR13530734	F8	PMM port 1	R2A, 25	47
F8_7S_10B	*Pseudomonas fulva*	JAFDRI000000000	SRR13530733	F8	Lab 3 overhead	R2A, 25	45
F8_7S_11B	*Pseudomonas fulva*	JAFDRJ000000000	SRR13530732	F8	Lab 3 overhead	R2A, 25	44
F8_7S_1B	*Pseudomonas fulva*	JAFDRR000000000	SRR13530723	F8	Lab 3 overhead	R2A, 25	46
F8_7S_1P	*Pseudomonas fulva*	JAFDRS000000000	SRR13530722	F8	Lab 3 overhead	BA, 37	43
F8_7S_2B	*Pseudomonas fulva*	JAFDRT000000000	SRR13530721	F8	Lab 3 overhead	R2A, 25	46
F8_7S_2P	*Pseudomonas fulva*	JAFDRU000000000	SRR13530720	F8	Lab 3 overhead	BA, 37	48
F8_7S_3B	*Pseudomonas fulva*	JAFDRV000000000	SRR13530719	F8	Lab 3 overhead	R2A, 25	43
F8_7S_3P	*Pseudomonas fulva*	JAFDRW000000000	SRR13530718	F8	Lab 3 overhead	BA, 37	47
F8_7S_6B	*Pseudomonas fulva*	JAFDRZ000000000	SRR13530714	F8	Lab 3 overhead	R2A, 25	48
F8_7S_8B	*Pseudomonas fulva*	JAFDSC000000000	SRR13530711	F8	Lab 3 overhead	R2A, 25	45
F8_7S_9B	*Pseudomonas fulva*	JAFDSD000000000	SRR13530710	F8	Lab 3 overhead	R2A, 25	43
F8_8S_13B	*Pseudomonas fulva*	JAFDSG000000000	SRR13530707	F8	Crew quarters	R2A, 25	48
F8_8S_1B	*Pseudomonas fulva*	JAFDSH000000000	SRR13530706	F8	Crew quarters	R2A, 25	45

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F8_8S_2B	*Pseudomonas fulva*	JAFDSI000000000	SRR13530705	F8	Crew quarters	R2A, 25	47
F8_8S_3B	*Pseudomonas fulva*	JAFDSK000000000	SRR13530702	F8	Crew quarters	R2A, 25	49
F8_8S_7P	*Pseudomonas fulva*	JAFDSM000000000	SRR13530700	F8	Crew quarters	BA, 37	48
F8_8S_8P	*Pseudomonas fulva*	JAFDSN000000000	SRR13530699	F8	Crew quarters	BA, 37	50
F8_8S_9P	*Pseudomonas fulva*	JAFDSO000000000	SRR13530698	F8	Crew quarters	BA, 37	44
F6_4S_P_1A	*Pseudomonas granadensis*	JAFDPY000000000	SRR13530773	F6	Dining table	BA, 37	32
F6_4S_P_1B	*Pseudomonas granadensis*	JAFDPZ000000000	SRR13530772	F6	Dining table	BA, 37	40
F6_4S_P_1C	*Pseudomonas granadensis*	JAFDQA000000000	SRR13530771	F6	Dining table	BA, 37	36
F6_4S_P_2	*Pseudomonas granadensis*	JAFDQB000000000	SRR13530769	F6	Dining table	BA, 37	36
F6_4S_P_5C	*Pseudomonas granadensis*	JAFDQC000000000	SRR13530768	F6	Dining table	BA, 37	35
F6_7S_P_1	*Staphylococcus capitis*	JAFDQE000000000	SRR13530766	F6	Lab 3 overhead	BA, 37	23
F6_7S_P_4	*Staphylococcus capitis*	JAFDQG000000000	SRR13530764	F6	Lab 3 overhead	BA, 37	16
F5_7S_P12B	*Staphylococcus caprae*	JAFDPE000000000	SRR13530726	F5	Lab 3 overhead	BA, 37	12
F6_3S_P_7	*Staphylococcus epidermidis*	JAFDPX000000000	SRR13530774	F6	ARED	BA, 37	32
F5_7S_P10A	*Staphylococcus saprophyticus*	JAFDOY000000000	SRR13530792	F5	Lab 3 overhead	BA, 37	27

Sample name	Bacterial species	WGS accession no.	SRA accession no.	Flight no.	Location[a]	Medium, temp (°C)[b]	N co
F5_7S_P11B	*Staphylococcus saprophyticus*	JAFDPB000000000	SRR13530759	F5	Lab 3 overhead	BA, 37	26
F5_7S_P12A	*Staphylococcus saprophyticus*	JAFDPD000000000	SRR13530737	F5	Lab 3 overhead	BA, 37	27
F5_7S_P13	*Staphylococcus saprophyticus*	JAFDPF000000000	SRR13530715	F5	Lab 3 overhead	BA, 37	24
F5_7S_P5A	*Staphylococcus saprophyticus*	JAFDPN000000000	SRR13530785	F5	Lab 3 overhead	BA, 37	27
F5_7S_P5B	*Staphylococcus saprophyticus*	JAFDPO000000000	SRR13530784	F5	Lab 3 overhead	BA, 37	27
F5_7S_P5C	*Staphylococcus saprophyticus*	JAFDPP000000000	SRR13530783	F5	Lab 3 overhead	BA, 37	25
F6_7S_P_5	*Staphylococcus saprophyticus*	JAFDQH000000000	SRR13530763	F6	Lab 3 overhead	BA, 37	23
F5_7S_P4	*Staphylococcus warneri*	JAFDPM000000000	SRR13530786	F5	Lab 3 overhead	BA, 37	26

[a]WHC, waste and hygiene compartment; ARED, advanced resistive exercise device; PMM, permanent multipurpose module.
[b]BA, blood agar.

Four species from the phylum *Actinobacteria* were isolated, from the genera *Pseudoclavibacter*, *Kocuria*, and *Micrococcus*, all of which are common in both environmental and human microbiomes (2–5). *Kocuria palustris* and *Micrococcus luteus* are opportunistic pathogens (5, 6). *M. luteus* can survive in a dormant state under extreme oligotrophic conditions (7) and has increased growth and increased biomass yield in microgravity (8).

Multiple coagulase-negative staphylococci (CoNS) were also isolated. CoNS are normal components of human skin flora (9) and are often resistant to antibiotics because of their ability to form biofilms (10); all CoNS in this report are

documented opportunistic pathogens (11–15).

Three species from the phylum *Proteobacteria* were identified. *Acinetobacter pittii* is a less common nosocomial pathogen that causes pneumonia and meningitis in intensive care patients (16), *Pseudomonas fulva* is a commensal plant endophyte (17) that can also infect immunocompromised patients (18, 19), and *Pseudomonas granadensis* is a recently discovered soil bacterium (20).

Two other species in this report that are not associated with the human microbiome are *Cytobacillus horneckiae*, a Gram-positive UV-resistant endospore-former that was isolated from a clean room at the Kennedy Space Center (21), and *Methylobacterium organophilum*, a facultative methylotroph (22).

All strains reported here were collected aboard the ISS over the course of five flight missions between June 2017 and December 2018 (Table 1). Premoistened polyester wipes were used to collect samples from eight predetermined surfaces during each flight (see Table 1 for locations). After transport to Earth, the wipes were agitated in sterile phosphate-buffered saline, and the buffer was concentrated with an InnovaPrep CP-150 concentrator. Concentrates were plated onto Reasoner's 2A (R2A) agar (25°C for 7 days), potato dextrose agar (PDA) (25°C for 7 days), and blood agar (37°C for 2 days) using appropriate concentrations for microbial isolation. Isolated colonies were restreaked on tryptic soy agar (TSA) (25°C), and genomic DNA was extracted using the ZymoBIOMICS DNA MagBead kit according to the manufacturer's instructions. Libraries for whole-genome shotgun sequencing (WGS) were prepared using the Illumina Nextera DNA Flex library preparation kit as in previous studies (23) and were sequenced using the NovaSeq 6000 S4 flow cell paired-end 2 × 150-bp platform.

Sequencing reads were quality filtered and trimmed, and adapter sequences were removed, using FastQC v0.11.7 (24) and fastp v0.20.0 (25). Scaffolds were assembled with SPAdes v3.11.1 (26). QUAST v5.0.2 (27) was used to determine assembly quality, including the number of contigs, genome size, and N_{50} value. Default settings were used for all steps except for fastp, which included 512 adapters screening. OrthoANIu (28) was used to confirm the species identity for each strain against the species type strain sequence, with a minimum average nucleotide identity of 95% for identification. Genomes were annotated using the NCBI Prokaryotic Genome Annotation Pipeline (29).

Data availability.

The WGS data and raw data have been deposited in GenBank under the BioProject accession number PRJNA690512. This project has also been deposited in the NASA GeneLab system (30) under the project number GLDS-361 . The versions described in this paper are the first versions.

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Associated Data

This section collects any data citations, data availability statements, or supplementary materials included in this article.

Data Availability Statement

The WGS data and raw data have been deposited in GenBank under the BioProject accession number PRJNA690512. This project has also been deposited in the NASA GeneLab system (30) under the project number GLDS-361 . The versions described in this paper are the first versions.